Claims

1. Method for determining hybridization on a microarray, comprising:

   (a) providing a microarray with a plurality of probes;

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   (b) conducting *in situ* fractionation of hybridized target in at least one probe of the microarray by means of at least one wash with a defined stringency;

   (c) collecting labelling intensity data at or after the *in situ* fractionation with a defined stringency;

   (d) repeating steps (a) and (b), wherein in a subsequent cycle the defined
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   stringency is increased;

   (e) generating a set of data corresponding to at least the stringency and the respective labelling intensity data obtained by each cycle for said cycles according to step (c); and

   (f) analyzing the set of data for determining hybridization in at least one probe.

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2. Method according to claim 1, wherein the labelling intensity data is fluorescent intensity data.

3. Method according to claim 1, wherein step (a) comprises providing a DNA chip.
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4. Method according to claim 1 or 3, wherein step (e) comprises generating a fractionation curve.

5. Method according to claim 4, wherein based on characteristic features of the
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fractionation curve, unreliable data is filtered and eliminated from subsequent analyses.

6. Method according to claim 5, wherein the characteristic features comprise transition stringency.
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7. Method according to claim 5, wherein the characteristic features comprise correlation between transition stringency and a calculated temperature of the probe to detect cross-hybridisation.

8. Method according any of the preceding claims, wherein steps (a) to (f) are conducted for a plurality of probes or all probes of said microarray in order to identify probes that produce specific hybridization signals.

9. Method according to any of the preceding claims, with further steps or modified steps as derivable from the remaining specification.

10. Computer program product comprising program code means stored on a computer readable medium for performing the computable part of the method of any of the preceding claims, wherein said program product is capable of being executed by a computer.

11. Computer program product comprising program code means stored on a computer readable medium for performing the computable part of the method of any of the preceding claims, wherein said program product is run on a computer.

12. System for determining hybridization on a microarray, particularly for performing the method of any of claims 1 - 9, comprising:

    (a) a microarray with a plurality of probes;

    (b) means for repeatedly conducting *in situ* fractionation of hybridized target in at least one probe of the microarray by means of at least one wash with a defined stringency;

    (c) means for repeatedly collecting fluorescent intensity data at or after the *in situ* fractionation with a defined stringency;

    (d) means for generating a set of data corresponding to at least the stringency and the respective fluorescent intensity data obtained by each cycle for said cycles according to step (c); and

    (e) means for analyzing the set of data for determining hybridization in at least one probe.

13. System according to claim 12, wherein the microarray is a DNA chip.

14. System according to claim 12 or 13, wherein a computer is provided to generate a fractionation curve.

15. System according to claim 14, wherein filter means and/or analyzing means are provided for analyzing said fractionation curve in order to filter out unreliable data.

16. System according to any of claims 11 - 14, with further means or modified means as derivable from the remaining specification.

17. Use of a method according to any of claims 1-9, a computer program product according to claim 10 or 11, and/or a system according to any of claims 12 - 16 for identifying probes on DNA-chips that produce specific hybridization signals in DNA-chip expression profiling approaches.

18. A method of producing a pharmaceutical composition comprising formulating the compound identified, refined or modified by the method of any of claims 1 - 9, a computer program product according to claim 10 or 11, and/or a system according to any of claims 12 - 16, with a pharmaceutically active carrier or diluent.

19. Compound identified, refined or modified by the method of any of claims 1 - 9, a computer program product according to claim 10 or 11, and/or a system according to any of claims 12 - 16, with a pharmaceutically active carrier or diluent.